K083846

510(K) SUMMARY

SEP - 1 2009

Cystic Fibrosis 39 kit v2

This summary of 510(k) safety and effectiveness information is being submitted in accordance with the requirement of 21 CFR 807.92.

510(k) Number:
k083846

Purpose for Submission:
New Device.

Measurand:
CFTR (cystic Fibrosis transmembrane conductance regulator) gene from human blood specimens

Type of Test:
Qualitative nucleic acid multiplex test.

Applicant:
Luminex Molecular Diagnostics Inc.
439 University Ave.
Toronto, ON M5G 1Y8 Canada
Tel: 416.593.4323 x374
Fax: 416.593.1001
Contact person: Gloria Lee

Proprietary and Established Names:
xTAG® Cystic Fibrosis 39 kit v2

Regulatory Information:

1. Regulation Section:
21 CFR 866.5900, CFTR (cystic fibrosis transmembrane conductance regulator) gene mutation detection system

2. Classification:
Class II

3. Product Code:
NUA

4. Panel:
Immunology (82)

Intended Use:
The xTAG® Cystic Fibrosis 39 kit v2 is a device used to simultaneously detect and identify a panel of mutations and variants in the cystic fibrosis transmembrane conductance regulator (CFTR) gene in human blood specimens. The panel includes mutations and variants currently recommended by the American College of Medical Genetics and American College of Obstetricians and Gynecologists (ACMG/ACOG), plus some of the worlds most common and North American-prevalent mutations. The xTAG Cystic Fibrosis 39 kit v2 is a qualitative genotyping test which provides information intended to be used for carrier testing in adults of reproductive age, as an aid in newborn screening, and in confirmatory diagnostic testing in newborns and children.

The kit is not indicated for use in fetal diagnostic or pre-implantation testing. This kit is also not indicated for stand-alone diagnostic purposes.

Mutations (asterisk denotes ACMG/ACOG panel) and 4 variants (variants italized) included in the xTAG CFTR 39 kit v2

ΔF508*	1717-1G>A*	W1282X*	2307insA
ΔI507*	R560T*	1078delT	Y1092X
G542X*	R553X*	394delTT	M1101K
G85E*	G551D*	Y122X	S1255X
R117H*	1898+1G>A*	R347H	3876delA
621+1G>T*	2184delA*	V520F	3905insT
711+1G>T*	2789+5G>A*	A559T	*5/7/9T*
N1303K*	3120+1G>A*	S549N	*F508C*
R334W*	R1162X*	S549R	*I507V*
R347P*	3659delC*	1898+5G>T	*I506V*
A455E*	3849+10kbC>T*	2183AA>G	

Indication(s) for use: The xTAG Cystic Fibrosis 39 kit v2 is a genotyping test indicated in adults for detecting mutations in the CFTR gene and in newborns and children as an aid in the diagnosis of suspected cystic fibrosis.

Special conditions for use statement(s):
The kit is not indicated for use in fetal diagnostic or pre-implantation testing. This kit is also not indicated for stand-alone diagnostic purposes.

Special instrument requirements:
Luminex 100 or 200 instrument

Device Description:
The xTAG CFTR 39 kit v2 includes the following components:

- PCR Primer Mix v2 including dNTPs designed to simultaneously produce 23 amplimers of the CFTR gene (24 in the presence of CFTR del 2, 3).
- ASPE Mix A v2 including dNTPs contains primers designed to hybridize to either wild-type or mutant alleles with proprietary sequences at their 5' ends designed to specifically hybridize to complementary sequences coupled to a given bead population in Bead Mix A.
- Bead Mix A v2 contains spectrally distinguishable populations of polystyrene beads internally dyed with red and infrared fluorochromes coupled to proprietary DNA sequences designed to specifically hybridize to complementary sequences on the ASPE primers in ASPE Mix A v2.
- 10X Buffer

- Platinum® TFI DNA Polymerase
- Platinum® TFI Reaction Buffer
- TFI $MgCl_2$
- Shrimp Alkaline Phosphatase
- Exonuclease I
- Strepavidin-Phycoerythrin Conjugate
- xTAG Data Analysis Software (TDAS) CFTR

Substantial Equivalence Information:

1. Predicate device name(s):
xTAG® Cystic Fibrosis Kit

2. Predicate 510(k) number(s):
k043011, k060627

3. Comparison with predicate:

Parameter	xTAG Cystic Fibrosis 39 kit v2	xTAG Cystic Fibrosis Kit
Intended Use	The xTAG Cystic Fibrosis 39 kit v2 is a device used to simultaneously detect and identify a panel of mutations and variants in the cystic fibrosis transmembrane conductance regulator (CFTR) gene in human blood specimens. The panel includes mutations and variants currently recommended by the American College of Medical Genetics and American College of Obstetricians and Gynecologists (ACMG/ACOG), plus some of the worlds most common and North American-prevalent mutations.	The xTAG Cystic Fibrosis Kit is a device used to simultaneously detect and identify a panel of mutations and variants in the cystic fibrosis transmembrane conductance regulator (CFTR) gene in human blood specimens. The panel includes mutations and variants currently recommended by the American College of Medical Genetics and American College of Obstetricians and Gynecologists (ACMG/ACOG), plus some of the worlds most common and North American-prevalent mutations.
Indications for Use	The xTAG Cystic Fibrosis 39 kit v2 is a qualitative genotyping test which provides information intended to be used for carrier testing in adults of reproductive age, as an aid in newborn screening, and in confirmatory diagnostic testing in newborns and children.	The xTAG Cystic Fibrosis Kit is a qualitative genotyping test which provides information intended to be used for carrier testing in adults of reproductive age, as an aid in newborn screening, and in confirmatory diagnostic testing in newborns and children.
Contra-Indications	The kit is not indicated for use in fetal diagnostic or pre-implantation testing. This kit is also not indicated for stand-alone diagnostic purposes.	The kit is not indicated for use in fetal diagnostic or pre-implantation testing. This kit is also not indicated for stand-alone diagnostic purposes.
Type of Test	Multiplex PCR followed by multiplex allele specific primer extension for genotyping, hybridized to multiplex fluorescent microparticles, detected by flow cytometry.	Multiplex PCR followed by multiplex allele specific primer extension for genotyping, hybridized to multiplex fluorescent microparticles, detected by flow cytometry.
Product Description	Tests for 39 mutations and 4 variants in the CFTR gene (23 of which are recommended by ACMG/ ACOG). The mutations and variants are the same as those tested for by the predicate device.	Tests for 39 mutations and 4 variants in the CFTR gene (23 of which are recommended by ACMG/ ACOG).

Specimen Type	Peripheral human whole blood.				Peripheral human whole blood.			
Instrument System	Luminex 100 or 200 IS				Luminex 100 or 200 IS			
Software	TDAS CFTR contains 1 template to detect for mutations. Software masking function where user can chose to display results for only the ACMG / ACOG 23 mutations or the full panel of mutations.				TDAS CF-I contains 1 template to detect for 39 mutations and 4 variants.			
Mutations Detected	ΔF508	1717-1G>A	W1282X	2307insA	ΔF508	1717-1G>A	W1282X	2307insA
	Δ1507	R560T	1078delT	Y1092X	Δ1507	R560T	1078delT	Y1092X
	G542X	R553X	394delTT	M1101K	G542X	R553X	394delTT	M1101K
	G85E	G551D	Y122X	S1255X	G85E	G551D	Y122X	S1255X
	R117H	1898+1G>A	R347H	3876delA	R117H	1898+1G>A	R347H	3876delA
	621+1G>T	2184delA	V520F	3905insT	621+1G>T	2184delA	V520F	3905insT
	711+1G>T	2789+5G>A	A559T	5/7/9T	711+1G>T	2789+5G>A	A559T	5/7/9T
	N1303K	3120+1G>A	S549N	F508C	N1303K	3120+1G>A	S549N	F508C
	R334W	R1162X	S549R	I507V	R334W	R1162X	S549R	I507V
	R347P	3659delC	1898+5G>T	I506V	R347P	3659delC	1898+5G>T	I506V
	A455E	3849+10kbC>T	2183AA>G		A455E	3849+10kbC>T	2183AA>G	

Standard/Guidance Document Referenced (if applicable):

- American College of Medical Genetics (ACMG) / American College of Obstetricians and Gynecologists Technical Standards and Guidelines for CFTR Mutation Testing and Standards and Guidelines for Clinical Genetic Laboratories
- Cystic Fibrosis Foundation / Center for Disease Control Recommendations on Newborn Screening for CF
- FDA Class II Special Controls Guidance: Quality Control Material for Cystic Fibrosis Nucleic Acid Assays (Jan 2007)
- FDA Class II Special Controls Guidance: CFTR Gene Mutation Detection Systems (Oct 2005)
- CDRH Draft Guidance on Multiplex Tests for Heritable DNA Markers, Mutations and Expression Patterns (Feb 2003)
- CDRH Draft Guidance on Statistical Guidance on Reporting Results from Studies Evaluating Diagnostic Tests (Mar 2003)
- CDRH Guidance for the Content of Pre-Market Submission for Software Contained in Medical Devices (May 1998)
- CDRH Guidance on General Principles of Software Validation (Jan 2002)
- CDRH Guidance on Format for Traditional and Abbreviated 510ks (Aug 2005)
- MM01-A2: Molecular Diagnostic Methods for Genetic Diseases
- MM13-PE: Collection, Transport, Preparation, and Storage of Specimens for Molecular Methods
- MM17-A: Verification and Validation of Multiplex Nucleic Acid Assays
- EP05-A2: Evaluation of Precision Performance of Clinical Chemistry Devices
- EP07-A2E: Interference Testing in Clinical Chemistry
- EP12-A: User Protocol for Evaluation of Qualitative Test Performance
- EP17-A: Protocols for Determining Limits of Detection and Limits of Quantitation

Test Principle:

The xTAG CFTR 39 kit v2 incorporates multiplex Polymerase Chain Reaction (PCR) and multiplex Allele Specific Primer Extension (ASPE) with LMD's proprietary Universal Tag sorting system on the Luminex® 100 or 200 xMAP® platform.

The amplimer sizes range from 179 bp to 465 bp. A multiplex PCR reaction is carried out under optimized conditions. Each sample then undergoes a multiplex allele specific primer extension (ASPE) reaction where an aliquot of the PCR product is run through ASPE A reaction. The ASPE step allows for detection of each allele (wild-type or mutant) of a

given locus using an allele-specific probe (ASP) which contains a unique DNA sequence (tag) at its 5' end. Each bi-allelic locus has two ASPs and each tri-allelic loci has 3 ASPs included in the ASPE Mix. For each ASP, the 3' end of the primer is a perfect match for its allele, but will have a 3' mismatch on any other allele. Both these ASPs however are tagged with a common tag at their 5' end. The DNA polymerase will only extend the primer when there is a perfect match on the 3' end, so that the primer is only extended if its target allele is present in the sample. Biotin-dCTP is incorporated into the extending chain if extension occurs.

For the hybridization reaction, the ASPE reaction product is added directly to microwells containing aliquots of the Bead Mix A v2. Each coupled bead is spectrally distinguishable from the other coupled beads in a given bead mix. A fluorescent reporter molecule (streptavidin-phycoerythrin) is bound to the biotin on the extended primers. Each tagged primer hybridizes only to its unique anti-tag complement; therefore, each colored bead represents a specific allele, through the bead/anti-tag/tagged primer association. The beads are then analyzed by the Luminex xMAP instrument. The Luminex instrument contains two lasers: one identifies the color-coded bead, and the other identifies the presence or absence of extended allele specific primer through the phycoerythrin reporter. Thus, the genotype of that locus is identified by the presence of phycoerythrin signal attached to one or both ASPs.

For each sample analyzed by the xTAG Cystic Fibrosis 39 kit v2, an output file containing MFI signals from the Luminex instrument is generated. The proprietary software component of this product analyzes this output data file to provide a final qualitative genotype for the sample. The user must select between 2 options for the final output prior to running the assay:

Option 1: Full Panel (39 mutations/deletions + 4 variants).
Option 2: ACMG/ACOG panel (23 mutations and deletions).

Performance Characteristics (if/when applicable):

Clinical Performance Characteristics:
a) Method Comparison Studies / Accuracy:

Accuracy of the xTAG CFTR 39 kit v2 was assessed through evaluation of samples representing all alleles (mutations and polymorphisms) probed by the assay. The majority of samples consisted of left-over, anonymized, banked whole-blood specimens. These specimens were supplemented with genomic DNAs from EBV-transformed lymphoid cell lines, and several custom-designed plasmids engineered to contain 1-2 CFTR mutations each. Archived clinical genomic DNA samples were obtained from a variety of sources.

The FDA cleared xTAG Cystic Fibrosis Kit (k043011 and k060627) was used as the comparator for all clinical specimens.

Table 1. Summary of Accuracy Study Results for the xTAG CFTR 39 kit v2

					Before allowable re-run					After allowable re-run				
Exon or Intron	Mutations	Number of independent clinical samples tested, per mutation	Number of Cell Lines Tested, per mutation	Number of Plasmids Tested, per mutation	Total no. repeats due to mis-calls	Total no. of repeats due to no-calls	% Accuracy prior to repeats	LB of 95% CI * ‡	UB of 95% CI * ‡	Total no. repeats due to mis-calls	Total no. repeats due to no-calls	Final % Accuracy (after Repeats)	LB of 95% CI * ‡	UB of 95% CI * ‡
	G85E #	2	0	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
Exon 3	394delTT	2	0	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
	R117H #	36	0	0	0	0	100.00	90.51	100.00	0	0	100.00	90.51	100.00
	Y122X	1	1	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
Exon 4	621+1G>T #	6	0	0	0	0	100.00	54.07	100.00	0	0	100.00	54.07	100.00
Exon 5	711+1G>T #	3	0	0	0	0	100.00	29.24	100.00	0	0	100.00	29.24	100.00
	1078delT	3	0	0	0	0	100.00	29.24	100.00	0	0	100.00	29.24	100.00
	R334W #	3	0	0	0	0	100.00	29.24	100.00	0	0	100.00	29.24	100.00
	R347Pmut #	6	0	0	0	0	100.00	54.07	100.00	0	0	100.00	54.07	100.00
Exon 7	R347Hmut	3	1	0	0	0	100.00	39.76	100.00	0	0	100.00	39.76	100.00
EXON 9	A455E #	3	0	0	0	0	100.00	29.24	100.00	0	0	100.00	29.24	100.00
	dI507mut #	9	0	0	0	0	100.00	66.37	100.00	0	0	100.00	66.37	100.00
	dF508mut #	162	1	0	0	0	100.00	97.87	100.00	0	0	100.00	97.87	100.00
Exon 10	V520F	2	0	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
	1717-1G>A #	5	0	0	0	0	100.00	47.82	100.00	0	0	100.00	47.82	100.00
	G542X #	13	0	0	0	0	100.00	75.29	100.00	0	0	100.00	75.29	100.00
	S549N	1	1	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
	S549R	2	1	0	0	0	100.00	47.82	100.00	0	0	100.00	47.82	100.00
	G551D #	12	0	0	0	0	100.00	73.54	100.00	0	0	100.00	73.54	100.00
	R553X #	7	0	0	0	0	100.00	59.04	100.00	0	0	100.00	59.04	100.00
	A559T	2	0	0	0	0	100.00	29.24	100.00	0	0	100.00	29.24	100.00
EXON 11	R560T #	4	0	0	0	0	100.00	39.76	100.00	0	0	100.00	39.76	100.00

Table 1 (continued). Summary of Accuracy Study Results for the xTAG CFTR 39 kit v2

					Before Allowable re-run					After allowable re-run				
Exon or Intron	Mutations	Number of independent clinical samples tested, per mutation ***	Number of Cell Lines Tested, per mutation	Number of Plasmids Tested, per mutation	Total no. repeats due to mis-calls	Total no. of repeats due to no calls	% Accuracy prior to repeats	LB of 95% CI ** 1	UB of 95% CI ** 1	Total no repeats due to mis-calls	Total no repeats due to no calls	Final % Accuracy (after Repeats)	LB of 95% CI ** 1	UB of 95% CI ** 1
	1898+1G>A #	2	0	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
Exon 12	1898+5G>T	0	0	2	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
	2183AA>G	2	0	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
	2184delA #	1	0	0	0	0	100.00	2.50	100.00	0	0	100.00	2.50	100.00
Exon 13	2307insA	3	0	0	0	0	100.00	29.24	100.00	0	0	100.00	29.24	100.00
EXON 14b	2789+5G>A #	5	0	0	0	0	100.00	47.82	100.00	0	0	100.00	47.82	100.00
Exon 16	3120+1G>A	7	0	0	0	0	100.00	59.04	100.00	0	0	100.00	59.04	100.00
	Y1092X-C>G	0	0	2	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
	Y1092X-C>A	2	0	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
Exon 17b	M1101K	0	2	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
	R1162X #	5	0	0	0	0	100.00	47.82	100.00	0	0	100.00	47.82	100.00
	3659delC #	4	0	0	0	0	100.00	39.76	100.00	0	0	100.00	39.76	100.00
Exon 19	S1255X(19)	4	0	0	0	0	100.00	39.76	100.00	0	0	100.00	39.76	100.00
INTRON 19	3849+10kb #	13	0	0	0	0	100.00	75.29	100.00	0	0	100.00	75.29	100.00
	S1255X(20)	4	0	0	0	0	100.00	39.76	100.00	0	0	100.00	39.76	100.00
	3876delA	1	1	0	0	0	100.00	29.24	100.00	0	0	100.00	29.24	100.00
	3905insT	2	0	0	0	0	100.00	15.81	100.00	0	0	100.00	15.81	100.00
EXON 20	W1282X #	8	0	0	0	0	100.00	63.06	100.00	0	0	100.00	63.06	100.00
EXON 21	N1303K #	6	0	0	0	0	100.00	54.07	100.00	0	0	100.00	54.07	100.00
EXON 10	I506V-var tg	3	0	0	0	0	100.00	47.82	100.00	0	0	100.00	47.82	100.00
EXON 10	I506V-variant	5	0	0	0	0	100.00	47.82	100.00	0	0	100.00	47.82	100.00
	I507V-variant	0	1	0	0	0	100.00	2.50	100.00	0	0	100.00	2.50	100.00
	F508C-variant	5	0	0	0	0	100.00	47.82	100.00	0	0	100.00	47.82	100.00

Exon or Intron	Basis for Overall Accuracy Calculation	Number of Independent Clinical Samples tested ***	Number of Cell Lines Tested	Number of Plasmids Tested	Number of Reruns	Overall Accuracy Per Sample Before Reruns	LB of 95% CI (before reruns)	UB of 95% CI (before reruns)	Overall Accuracy Per Sample After Reruns	LB of 95% CI (after reruns)	UB of 95% CI (after reruns)
all exons	Overall accuracy per sample.	319	8	not used in calculation	0	327/327 = 100.00	98.88%	100.00%	327/327 = 100.00	98.88%	100.00%

* N for CI calculations = total number of independent samples tested
‡ UB = Upper Bound, LB = Lower Bound, CI = Confidence Interval. Clopper-Pearson CI calculator provided by John C. Pezzullo (Kissimmee, Florida, USA) and is available at http://statpages.org/confint.html
# ACMG recommended mutations

Table 1 demonstrates 100% accuracy compared with the reference method.

Analytical Performance Characteristics:
a) Precision/Reproducibility:

A multi-centre, multi-operator, multi-lot, blinded study design was used to evaluate total variability of the xTAG Cystic Fibrosis 39 kit v2.

The reproducibility of the analytical (post-extraction) steps of the assay was evaluated at 3 external sites (Hartford Hospital, Connecticut, USA = site 1, Luminex Molecular Diagnostics Inc., Toronto, Canada = site 2, Hospital for Sick Kids, Toronto, Canada = site 3), using in order of preference and availability, purified genomic DNAs extracted from clinical (whole blood) samples, purified genomic DNA extracted from lymphoid cell lines, and/or plasmids. Each set of samples contained samples representing all mutations and variants probed by the xTAG Cystic Fibrosis 39 kit v2. There were 2 operators per site, each performing 1 run / day across 3 non-consecutive days (3 runs per operator or 6 runs per site). Within a given run, each assay point was run in duplicate. A total of three (3) assay lots were tested (1 lot / site).

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Table 2. Reproducibility for xTAG Cystic Fibrosis 39 kit v2 (between site and between operator)

			Operator - to - Operator											
			Site 1				Site 2				Site 3			
Sample	Genotype		†Op 1 N¥	Op 1 % corr ‡	Op2 N	Op2 % corr	Op1 N	Op1 % corr	Op2 N	Op2 % corr	Op1 N	Op1 % corr	Op2 N	Op2 % corr
1	711+1G>T	dF508	438	100	438	100	438	100	438	100	438	100	438	100
2	1717-1G>A	-	438	100	438	100	438	100	438	100	438	100	438	100
3	G542X	R117H	438	100	438	100	438	100	438	100	438	100	438	100
4	A455E	-	438	100	438	100	438	100	438	100	438	100	438	100
5	3659delC	-	438	100	438	100	438	100	438	100	438	100	438	100
6	R1162X	dF508	438	100	438	100	438	100	438	100	438	100	438	100
7	3849+10kbC>T	-	438	100	438	100	438	100	438	100	438	100	438	100
8	W1282X	-	438	100	438	100	438	100	438	100	438	100	438	100
9	1078delT	dF508	438	100	438	100	438	100	438	100	438	100	438	100
10	A559T	-	438	100	438	100	438	100	438	100	438	100	438	100
11	S549N	-	438	100	438	100	438	100	438	100	438	100	438	100
12	G551D	R347P	438	100	438	100	438	100	438	100	438	100	438	100
13	3905insT	-	438	100	438	100	438	100	438	100	438	100	438	100
14	R560T	dF508	438	100	438	100	438	100	438	100	438	100	438	100
15	394delTT	-	438	100	438	100	438	100	438	100	438	100	438	100
16	R553X	-	438	100	438	100	438	100	438	100	438	100	438	100
17	2184delA	-	438	100	438	100	438	100	438	100	438	100	438	100
18	1898+1G>A	dF508	438	100	438	100	438	100	438	100	438	100	438	100
19	Y1092X-C>A	dF508	438	100	438	100	438	100	438	100	438	100	438	100
20	2183AA>G	-	438	100	438	100	438	100	438	100	438	100	438	100
21	V520F	3120+1G>A	438	100	438	100	438	100	438	100	438	100	438	100

Table 2 (continued). Reproducibility for xTAG Cystic Fibrosis 39 kit v2 (between site and between operator)

			Operator - to - Operator											
			Site 1				Site 2				Site 3			
Sample	Genotype		†Op 1 N¥	Op 1 % corr ‡	Op2 N	Op2 % corr	Op1 N	Op1 % corr	Op2 N	Op2 % corr	Op1 N	Op1 % corr	Op2 N	Op % c
22	R334W	-	438	100	438	100	438	100	438	100	438	100	438	10
23	2789+5G>A	-	438	100	438	100	438	100	438	100	438	100	438	10
24	612+1 G>A	-	438	100	438	100	438	100	438	100	438	100	438	10
25	dI507	-	438	100	438	100	438	100	438	100	438	100	438	10
26	dF508 (+ F508C variant)	-	439*	100	442**	100	438***	100	438***	100	438***	100	439*	100.
27	G85E	-	438	100	438	100	438	100	438	100	438	100	438	10
28	N1303K	-	438	100	438	100	438	100	438	100	438	100	438	10
29	M1101K	M1101K	438	100	438	100	438	100	438	100	438	100	438	10
30	Y122X	R1158X	438	100	438	100	438	100	438	100	438	100	438	10
31	R347H	-	438	100	438	100	438	100	438	100	438	100	438	10
32	3876delA	-	438	100	438	100	438	100	438	100	438	100	438	10
33	S549R	-	438	100	438	100	438	100	438	100	438	100	438	10
34	dF508	-	438	100	438	100	438	100	438	100	438	100	437	99.
35 36	dF508 (+ I506V variant)	V520F	18	100	18	100	18	100	18	100	18	100	18	10
37	1898+5G>T	-	6	100	6	100	6	100	6	100	6	100	6	10
38	2307insA	2055del9>A	12	91.67	12	83.33	12	100	12	100	12	100	12	10
39	3791delC	-	12	100	12	100	12	100	12	100	12	100	12	10
40	Y1092X-C>G	-	6	100	6	100	6	100	6	100	6	100	6	10
41	S1255X (ex.19)	-	6	100	6	100	6	100	6	100	6	100	6	10
42	S1255X (ex.20)	W1282X	12	100	12	100	12	100	12	100	12	100	12	10

* Site 1 = Hartford Hospital, Connecticut. USA; Site 2 = Luminex Molecular Diagnostics, Toronto, Canada; Site 3 = Hospital for Sick Children, Toronto, Canada.
† Op = operator (1 or 2)
** N, number of calls
‡, % corr, percent correct
* Total Number of calls 438 + 1 = 439, because TDAS made one dF508 Mu D call (F508C variant was unmasked)
Total Number of calls 438 + 4 = 442, because TDAS made 4 dF508 Mu D calls (F508C variant was unmasked)
Total Number of calls = 438, because TDAS made all dF508 HET calls (F508C variant was masked)

Table 2 shows that the xTAG Cystic Fibrosis 39 kit v2 assay detected all 39 mutations, as well as normal (wild-type) alleles, with a precision of > 99.54% across 3 sites, between 6 operators (2 per site) and between reagent lots (a total of 3 lots, 1 lot per site). Sample 34 (Coriell genomic DNA) made a 'No Call' after an allowable rerun at Site 3 (operator 1) whereas sample 37 (plasmid) made 3 miscalls at Site 1 between 2 operators.

Reproducibility of detection of a compound heterozygote dF508 / F508C was also characterized in this study. Of the 36 replicates of sample 26 tes 30 generated a dF508 HET call and 6 generated a dF508 Mu D call. Both results are accurate when taking into consideration the definition of a Mu call (i.e. only the mutant allele is detected).

Table 3 Reproducibility of the xTAG Cystic Fibrosis 39 kit v2 (per allele)

			Over All 3 Sites											
			Before Allowable Re-Run						After Allowable Re-Run					
Panel	Genotype	Total # calls (All Sites)	Total No. Missed Calls	Total No. No-Calls	Total No. Correct Calls	% Agreement with Comparator	LB of 95% CI *	UB of 95% CI *	Total No. Missed Calls	Total No. No-Calls	Total No. Correct Calls	% Agreement with Comparator	LB of 95% CI *	UB of 95% CI *
A	G85E	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	394delTT	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	R117H	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	Y122X	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	621+1G>T	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	711+1G>T	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	1078delT	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	R334W	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	R347P	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	R347H	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	A455E	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	dI507	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	dF508	468	0	16	452	96.58	94.51	98.03	0	2	466	99.57	98.46	99.95
A	V520F	72	0	0	72	100.00	95.01	100.00	0	0	72	100.00	95.01	100.00
A	1717-1G>A	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	G542X	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	S549N	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	S549R	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	G551D	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	R553X	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	A559T	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	R560T	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	1898+1G>A	36	0	1	35	97.22	85.47	99.93	0	0	36	100.00	90.26	100.00
A	1898+5G>T	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	2183AA>G	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	2184delA	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	2307insA	36	3	0	33	91.67	77.53	98.25	3	0	33	91.67	77.53	98.25

A	**2789+5G>A**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**3120+1G>A**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**Y1092X-C>G**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**Y1092X-C>A**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**M1101K**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**R1162X**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**3659delC**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**S1255X(19)**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**S1255X(20)**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**3849+10kb**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**3876delA**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**3905insT**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**W1282X**	72	0	0	72	100.00	73.54	100.00	0	0	72	100.00	73.54	100.00
A	**N1303K**	36	0	0	36	100.00	90.26	100.00	0	0	36	100.00	90.26	100.00
A	**Total WT Calls across all samples**	**54612**	**0**	**87**	**54525**	**99.841**	**99.804**	**99.872**	**0**	**0**	**54612**	**100.00**	**99.993**	**100.00**

* UB = Upper Bound, LB = Lower Bound, CI = Confidence Interval. Exact calculation (Clopper & Pearson (1934) Biometrika 26, 404-143.) Excel Macro from http://statpages.org/confing.html

The xTAG Cystic Fibrosis 39 kit v2 assay detects mutant / variant / wild-type alleles of the 39 loci assayed with reproducibility (after allowed reruns) of 91.67% for allele 2307insA, 99.57% for allele dF508mut, and 100.00% for the remaining alleles. For reproducibility testing of the 2307insA allele, a plasmid DNA was used (Table 2 and Table 3). In 3 out of 36 test points this sample was detected as a HET instead of a Mu D however, in the accuracy study (Table 1), all clinical samples representing the 2307insA mutation were correctly identified by this assay. Table 3 shows that across all alleles, the reproducibility of the assay is 100% after allowable re-runs.

.b) Traceability, Stability, Expected Values (controls, calibrators, or methods):
N/A

c) Detection Limit and range of assay:
Genomic DNA samples representing a subset of mutations in the CFTR 39 kit v2 test were assayed at the following concentrations: 300, 150, 50, 25, 12.5, 6.25, 3.125, 1.56, 0.78 and 0.39 ng/µL. Each sample was run in duplicate. Genomic DNA, extracted from whole blood, was used as a wild-type positive control in all runs. At each tested concentration, the data across all samples were pooled. The lowest concentration giving an apparent assay failure rate of ≤5% was considered an estimator of the LoD. The proposed assay lower bound (LB) was set at a concentration (C*) lying at or slightly above the estimated LoD. At concentration C*, 22 replicates of each of the 10 genomic DNAs were run along with 8 negative controls dispersed uniformly throughout the plate to determine the LoD. The Lower Bound and Upper Bound of the assay range was determined to be 2 ng/µL and 300 ng/µL, respectively. The LoD was determined to be 1.56 ng/µL.

d) Analytical Specificity / Interfering Sub stances:

An Interference study was conducted to examine the effects of potential interferents that might be expected to be found in whole blood samples (1500 µg/mL hemoglobin, 200 µg/mL bilirubin, and 30 mg/mL mixture of triglycerides). Eight whole blood samples were split into 6 parts each, and incubated either in the absence or presence of one of the 3 potential interferents, extracted and assayed with CFTR 39 kit v2. No difference was observed between the final qualitative calls made from the untreated vs treated samples. This study showed that none of the potential interferents commonly found in whole blood produced a significant inhibitory effect on the performance of the CFTR 39 kit v2.

e) Stability:
The expiration date for xTAG CFTR 39 kit v2 will be based on real-time stability testing.

f) Assay Cut-off:
N/A

DEPARTMENT OF HEALTH & HUMAN SERVICES Public Health Service

Food and Drug Administration
10903 New Hampshire Avenue
Document Mail Center - WO66-G609
Silver Spring, MD 20993-0002

Luminex Molecular Diagnostics, Inc.
c/o Gloria Lee
439 University Avenue, Suite 2000
Toronto, Ontario
Canada M5G 1Y8

SEP - 1 2009

Re: k083846
Trade/Device Name: xTAG™ Cystic Fibrosis 39 Kit v2
Regulation Number: 21 CFR 866.5900
Regulation name: CFTR (cystic fibrosis transmembrane conductance regulator) gene mutation detection system
Regulatory Class: Class II
Product Code: NUA
Dated: August 24, 2009
Received: August 25, 2009

Dear Ms. Lee:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must

comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820). This letter will allow you to begin marketing your device as described in your Section 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific advice for your device on our labeling regulation (21 CFR Parts 801 and 809), please contact the Office of *In Vitro* Diagnostic Device Evaluation and Safety at (301) 796-5450. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm for the CDRH's Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or (301) 796-7100 or at its Internet address http://www.fda.gov/cdrh/industry/support/index.html.

Sincerely yours,

Leena Philip

For Maria M. Chan, Ph.D.
Director
Division of Immunology and Hematology Devices
Office of In Vitro Diagnostic Device Evaluation and Safety
Center for Devices and Radiological Health

Enclosure

Indications for Use

510(k) Number: k083846

Device Name: xTAG® Cystic Fibrosis 39 kit v2

Indications For Use:

The xTAG® Cystic Fibrosis 39 kit v2 is a device used to simultaneously detect and identify a panel of mutations and variants in the cystic fibrosis transmembrane conductance regulator (CFTR) gene in human blood specimens. The panel includes mutations and variants currently recommended by the American College of Medical Genetics and American College of Obstetricians and Gynecologists (ACMG/ACOG) plus some of the world's most common and North American prevalent mutations. The xTAG® Cystic Fibrosis 39 kit v2 is a qualitative genotyping test which provides information intended to be used for carrier testing in adults of reproductive age, as an aid in newborn screening, and in confirmatory diagnostic testing in newborns and children.

The kit is not indicated for use in fetal diagnostic or pre-implantation testing. This kit is also not indicated for stand-alone diagnostic purposes.

Prescription Use X AND/OR Over-The-Counter Use _____
(Part 21 CFR 801 Subpart D) (21 CFR 807 Subpart C)

(PLEASE DO NOT WRITE BELOW THIS LINE-CONTINUE ON ANOTHER PAGE IF NEEDED)

Concurrence of CDRH, Office of In Vitro Diagnostic Devices (OIVD)

Division Sign-Off

Office of In Vitro Diagnostic
Device Evaluation and Safety

510(k) k083846